AD_____

Award Number: DAMD17-02-1-0115

TITLE: The Isolation and Characterization of Prostate Stem Cells

PRINCIPAL INVESTIGATOR: Sarah N. Salm, Ph.D.

CONTRACTING ORGANIZATION: New York University School of Medicine
New York, New York 10016

REPORT DATE: February 2003

TYPE OF REPORT: Annual Summary

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

20030616 099

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 074-0188

Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing this collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Washington Headquarters Services, Directorate for Information Operations and Reports, 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302, and to the Office of Management and Budget, Paperwork Reduction Project (0704-0188), Washington, DC 20503

1. AGENCY USE ONLY (Leave blank)	2. REPORT DATE	3. REPORT TYPE AND DATES COVERED
	February 2003	Annual Summary (15 Jan 02 - 14 Jan 03)

4. TITLE AND SUBTITLE
The Isolation and Characterization of Prostate Stem Cells

5. FUNDING NUMBERS
DAMD17-02-1-0115

6. AUTHOR(S) :
Sarah N. Salm, Ph.D.

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
New York University School of Medicine
New York, New York 10016

Email: salms01@popmail.med.nyu.edu

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)
U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSORING / MONITORING AGENCY REPORT NUMBER

11. SUPPLEMENTARY NOTES
Original contains color plates: All DTIC reproductions will be in black and white.

12a. DISTRIBUTION / AVAILABILITY STATEMENT
Approved for Public Release; Distribution Unlimited

12b. DISTRIBUTION CODE

13. Abstract ***(Maximum 200 Words)*** *(abstract should contain no proprietary or confidential information)*
Stem cells are of considerable importance in prostate cancer because of the theory that cancer cells represent the malignant counterparts of normal tissue stem cells. We have shown that murine prostatic stem cells reside in the proximal region of the prostatic ducts. The in vivo growth properties and proliferative potential of the proximal cells were examined in a tissue transplantation model and in an orthotopic model. Some in vitro characteristics of their growth have also been examined.Cells were isolated from the proximal and distal regions of the prostate and were embedded in collagen gels in the absence or presence of smooth muscle cells (SMC) or fetal mouse or rat urogenital mesenchyme (UGM). The collagen pellets were implanted under the renal capsule of athymic mice and left for 8 weeks, after which the grafts were removed and weighed. No significant difference was noted in grafts containing either proximal or distal cells. This result was unaffected by SMC or fetal mouse UGM. In the presence of fetal rat UGM, grafts containing proximal cells were13.8 times larger than those containing distal cells, indicating a high proliferative potential in the proximal cells compared with the distal cells.In the orthotopic model, prostatic cells expressing GFP were injected into either intact prostates of juvenile mice or into the involuted prostates of castrated adult mice that were then given androgens to stimulate prostatic regrowth. In both cases, the GFP-expressing cells engrafted into the growing prostates, demonstrating the regenerative ability of the injected cells.
Finally, to characterize the stem cells, in vitro growth assays were performed. Proximal cells were grown in vitro in the presence or absence of stem cell factor (SCF) and antibodies to transforming growth factor-beta (TGF-ß). SCF stimulates the growth of the proximal cells when endogenous TGF-ß is inhibited, suggesting that these cells express c-kit, an established stem cell marker in a number of other systems such as hematopoiesis.

14. SUBJECT TERMS:	15. NUMBER OF PAGES
prostatic stem cells, transplant	11
	16. PRICE CODE

17. SECURITY CLASSIFICATION OF REPORT	18. SECURITY CLASSIFICATION OF THIS PAGE	19. SECURITY CLASSIFICATION OF ABSTRACT	20. LIMITATION OF ABSTRACT
Unclassified	Unclassified	Unclassified	Unlimited

Table of Contents

Introduction

Stem cells are of considerable importance in prostate cancer because of the theory that cancer cells represent the malignant counterparts of normal tissue stem cells (1). Understanding and characterizing the prostatic stem cells is important for defining the cellular events that take place during normal growth of the organ and during the aberrant proliferation processes of benign prostatic hyperplasia (BPH) and prostatic carcinoma. Although conclusive evidence is lacking, it is generally assumed that the prostate stem cells are located in the basal epithelium of the organ (2). However, little is known about these stem cells, their numbers, their exact location in the prostate and their characteristics. Our laboratory has recently demonstrated that there is a population of slow-cycling cells in the proximal region of the prostate. These cells possess a high in vitro proliferative capacity and can reconstitute highly branched glandular ductal structures in collagen gels. These three features are indicative of stem cells and we therefore propose that this population of cells contains the prostatic stem cells (3). The major goal of this fellowship proposal is to identify and isolate the stem cells of the murine prostate and to study them both in vitro and in vivo. The information gained from this work will give insight into the mechanisms involved in prostatic carcinogenesis and may enable further development of new therapies aimed at preventing carcinogenesis or delaying progression of this disease.

Body

Two tasks were outlined for the first year of this work. First, I proposed to study the in vivo growth properties and proliferative potential of putative prostatic stem cells in a tissue transplantation model. Second, I proposed to study the in vivo reconstitution potential of the putative stem cells in an orthotopic model.

Task 1. To study the in vivo growth properties and proliferative potential of putative prostatic stem cells in a tissue transplantation model.

Prostatic cells were isolated from the proximal and distal regions of the prostate. These cells were embedded in collagen gels in the presence or absence of either smooth muscle cells (SMC), or urogenital sinus mesenchyme (UGM) obtained from fetal mice or rats (1×10^5 prostate cells alone or mixed with 2.5×10^5 UGM cells, in 20 μl of collagen). The collagen pellets were then implanted under the renal capsule of athymic mice and allowed to develop for 6 weeks, after which the grafts were removed, weighed, fixed and examined by immunhistochemistry.

No significant difference was observed in the volume of grafts containing proximal cells alone ($119.3mm^3 \pm 40.9$) or distal cells alone ($92.4mm^3 \pm 30.6$). Combination of the prostate cells with SMC did not alter the growth of the grafts (not shown). Minimal growth was noted with SMC cells alone (not shown).

This experiment was repeated using UGM from 16-day old fetal mice. Although the grafts were larger when UGM was included with the prostate cells, there was no significant difference in weight between grafts containing distal prostate cells and UGM ($6.4mg \pm 2.1$) and those containing proximal cells and UGM ($12.3mg \pm 6.8$; $p<0.132$). However, in both cases the prostatic tissue was architecturally correct, with ducts having both basal and luminal epithelial layers surrounding a central lumen containing secretory products (Figure 1A and B).

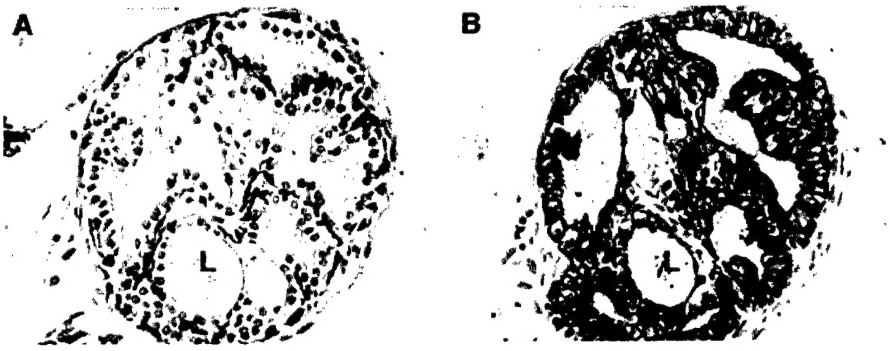

Figure 1A and B. Prostatic tissue in collagen grafts seeded with proximal cells ($1x10^5$) and fetal mouse UGM ($2.5x10^5$), after 6 weeks of incubation. A. Prostatic duct showing expression of cytokeratins 5 and 14 in basal cells. B. Prostatic duct showing expression of cytokeratins 8 and 18 in luminal cells. L is the central lumen containing prostatic secretions.

As Hayward et al (4) have noted that fetal rat UGM is more effective than fetal mouse UGM in this type of transplantation model, we repeated these experiments using UGM from 18-day-old fetal rats. There was a significant difference in the size of the grafts in kidneys receiving pellets with proximal cells compared with those receiving pellets containing distal cells (Figure 2).

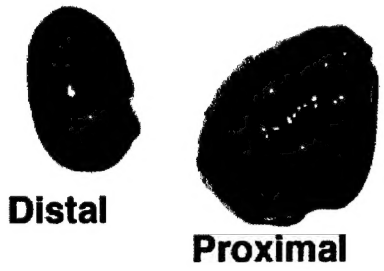

Figure 2. Gross morphology of kidneys containing sub-renal capsule grafts of collagen with UGM and either proximal or distal cells. Grafts contained either proximal or distal cells ($1x10^5$ cells/graft) and fetal rat UGM ($2.5x10^5$ cells/graft).

Measurement of the wet weights of grafts revealed that those containing proximal cells were 13.8 times larger than those containing distal cells (Figure 3).

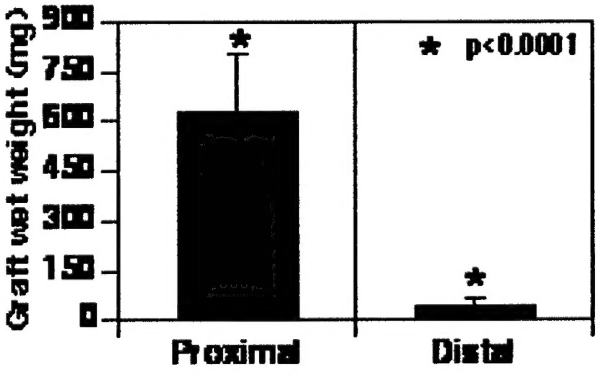

Figure 3. Wet weight of sub-renal capsule collagen grafts. Grafts contained either proximal or distal cells ($1x10^5$ cells/graft) and fetal rat UGM ($2.5x10^5$ cells/graft).

Control pellets containing UGM cells alone gave rise to minimal tissue formation (not shown). In addition, the size of the grafts containing prostate cells in combination with fetal rat UGM cells was significantly larger than those obtained using either proximal or distal prostate cells alone, or proximal or distal cells in combination with SMC or fetal mouse UGM. Examination of these grafts for ductal

architecture and cytokeratin expression revealed that they were architecturally correct and that both basal and luminal cytokeratins were expressed in the epithelial layer which surrounded a central lumen, as depicted in Figure 1.

These data indicate that in an in vivo transplantation model the proximal cells are highly proliferative and are capable of forming significant amounts of prostatic tissue under the renal capsule. The distal cells do not form equivalent amounts of tissue, suggesting that although they show some proliferative potential, it is significantly lower than that displayed by the proximal cells. The significantly increased tissue regeneration by the proximal cells fulfills a stem cell criterion, confirming our hypothesis that the prostate stem cells reside primarily in this region of the ducts.

Task 2. To study the in vivo reconstitution potential of the putative stem cells in an orthotopic model.

To refine the optimal conditions for using the orthotopic model, I initially approached Task 2 using well characterized basal and luminal cell lines (5,6) that have been transfected to express GFP. The remainder of task 2 will be completed in the second year of my fellowship as I decided it was scientifically necessary to start the in vitro characterization of the proximal cells (Task 3) during the current year.

GFP-transfected prostatic epithelial cell lines (basal and luminal) were injected into either the intact prostates of 3 week old mice or into the involuted prostates of castrated 6 week old wild type mice. The prostate develops post-natally and I therefore expected that GFP-cells would incorporate and engraft into 3-week old prostates during their development. In the case of 6 week old (adult) mice, the prostate is fully developed and I therefore created a niche for potential GFP-cell engraftment by castrating the mice and allowing the prostate to involute, priming it for regrowth which would be induced by using androgens. Recipient castrated mice were cycled at least three times by removing and replacing androgens, to ensure incorporation and grafting of the injected GFP-cells and to induce proliferation of these cells once engrafted. Each involution/regrowth cycle takes a minimum of 20 days, which allows for 10 days for complete prostatic involution followed by 10 days for complete regrowth, as dictated by the literature (7). Prostates were then removed and examined immunohistochemically for the presence of GFP-expressing epithelial cells.

I found that both GFP-basal and GFP-luminal cells engrafted into involuted prostates induced to regrow in the presence of androgens. Since the basal cells are considered to contain a population of stem cells (4), we had expected some of these cells to engraft. However, luminal cells are considered to arise from basal cells (4) and their ability to engraft was somewhat unexpected. However, as we have recently shown that both the basal and the luminal compartments of the mouse prostate contain slow cycling cells (3), it is possible that putative prostatic stem cells may exist in both the basal and the luminal epithelium of the prostate. This is the first indication that luminal cells can engraft in vivo to repopulate a regenerating prostate. I also found that while between 35 and 50% of the prostates examined from the mice injected at 3 weeks contained GFP-expressing cells, 60 to 80% of the prostates from the castrated, androgen cycled mice contained GFP positive cells after 3 cycles of androgen removal and supplementation. These data indicate that using 6 week old castrated mice as recipients for intraprostatic inoculation of GFP cells is more successful than using immature intact mice.

Technically, it was extremely difficult to inject sufficient cells into the prostates of the 3-week old mice, which may account for the lower number of engrafted cells. Also, since these mice were intact

and did not undergo androgen cycling, it is possible that the stem cell niche may not have been induced. These mice received a single injection of GFP cells and the prostate was left to develop normally. Completion of Task 2 requires injection of proximal or distal cells from GFP mice and will be undertaken this year. The cells will be injected into the involuted prostates of 6 week old castrated mice, and the mice will then be subject to at least 3 cycles of androgen removal and supplementation, as indicated by the data above.

Task 3. To study the in vitro characteristics of prostatic stem cells.

Task 3 is aimed at a second strategy for identifying stem cells, namely defining cellular markers for the isolation and characterization of these cells. Several markers have been identified in other stem cell populations that make them phenotypically identifiable from more mature cells. In the hematopoietic system for example, progenitor cells express c-kit, which is used to separate them from more mature progeny (8-10). Completion of this task will identify cellular markers in the prostatic stem cells.

Prostatic cells were removed from the proximal and distal regions of the prostatic ducts and were seeded at $2x10^4$ cells/well into collagen-coated wells in a 96 well plate. The cells were suspended in a growth medium described by us previously (5,6). Wells were then also supplemented with stem cell factor (SCF; 100 ng/ml), a TGF-ß neutralizing antibody (20 µg/ml) or a combination of the SCF and the TGF-ß neutralizing antibody. Ablation of endogenous TGF-ß activity with a neutralizing antibody stimulated proximal cell growth by 53% while concurrent neutralization of the TGF-ß with addition of SCF results in a 64% increase in growth. SCF alone did not result in a significant increase in growth (Figure 4).

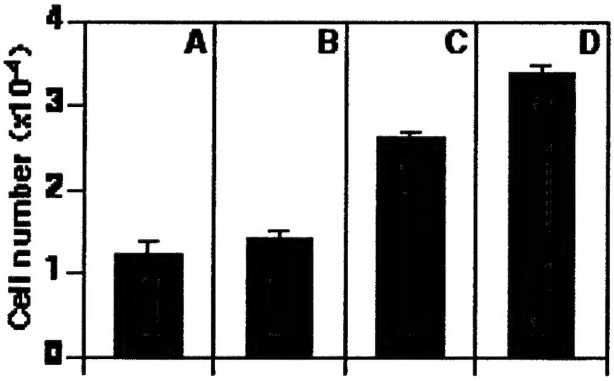

Figure 4. Proximal cells were seeded at $2x10^4$ cells in collagen-coated wells with factors. Cells were removed and counted after 6 days of culture. **A.** Control well. **B.** SCF (100 ng/ml). **C.** Anti-TGF-ß antibody (20 µg/ml). **D.** SCF + anti-TGF-ß antibody.

These results indicate that the proximal cells, which contain the putative stem cells, are stimulated by SCF in the absence of endogenous TGF-ß. SCF is the ligand for c-kit, which is a surface marker found on many primitive cells and stem cells, including hematopoietic, liver and spermatogonial stem cells (8-10) and the response of the proximal cells to this factor suggests that the prostatic stem cells also express c-kit. In the presence of TGF-ß, SCF has no significant effect on the growth of the cells, suggesting that prostatic homeostasis may be maintained by a balance between growth-stimulating factors such as SCF and growth-inhibitory factors such as TGF-ß.

Key Research Accomplishments

- We have successfully used a tissue transplantation model to demonstrate that prostate proximal cells can regenerate prostatic tissue under the renal capsule. We have shown that the reconstituted tissue contains functional basal and luminal epithelium as found in the adult prostate. We have

mastered the technically challenging process of isolating and preparing fetal rat UGM, which is needed for the success of this model.

- Using this model, we further demonstrated that the presence of fetal rat mesenchyme is required for optimum tissue regeneration, although even in the absence of mesenchymal cells, prostatic proximal cells are capable of regenerating functioning prostatic ducts.
- We have refined our intraprostatic transplantation model for stem cell identification using GFP-expressing prostate cell lines and have defined the optimal conditions for the proposed experiments using cells isolated from the proximal and distal regions of prostatic ducts. We have shown that inoculating cells into a young intact prostate is less effective for engraftment that inoculating them into adult, involuted prostates followed by cycling prostatic regrowth with the addition and removal of androgens.
- We have shown that growth of the proximal cells is stimulated by SCF, in the absence of endogenous TGF-ß. This suggests that these cells have c-kit, a cellular marker used to identify stem cells in other systems, such as hematopoiesis. It also implies that homeostasis of the stem cell compartment is maintained by a balance between growth-stimulating factors such as SCF and growth-inhibitory factors such as TGF-ß.

Reportable Outcomes

- **Conferences:**

  Abstracts submitted for The First Annual Stem Cell Wetlab/Conference in Providence, RI, April 8-11, 2003 (see appendix)
  a) **The location and characterization of murine prostatic stem cells.** Salm S, Tsujimura A, Koikawa Y, Takao T, Coetzee S, Moscatelli D, Sun T-T, Wilson EL.
  b) **The proximal region of murine prostatic ducts is enriched in prostate stem cells.** Ken Goto, Sarah Salm, Sandy Coetzee, Herbert Lepor, Ellen Shapiro, David Moscatelli and E. Lynette Wilson.

Conclusions

The work I have completed thus far has indicated that the murine prostatic stem cells lie within the proximal region of the prostatic ducts. These cells are slow cycling but are highly proliferative. I have used an in vivo model to demonstrate that cells removed from the proximal region of prostatic ducts are capable of regenerating complete, architecturally correct, functional prostatic ducts under the renal capsule. These data indicate that the proximal cells exhibit the most relevant features of stem cells, namely being able to self-renew and being able to reconstitute functional prostatic tissue in vivo. I have also refined a second in vivo model that will allow me to demonstrate how these cells function in situ within the stem cell niche of a murine prostate.

Finally I have begun to characterize the cells from the proximal region and have shown that the concurrent addition of SCF and neutralization of endogenous TGF-ß enhanced the growth of these cells. Further data gained from these experiments will indicate which cytokines enhance the proliferation of the stem cells in vitro and may be used to optimize culture conditions for future work. They will show whether the putative stem cells have a characteristic phenotype in terms of growth and differentiation and whether this phenotype can be maintained in culture.

References

1. Sell S, Pierce GB. 1994. Maturation arrest of stem cell differentiation is a common pathway for the cellular origin of teratocarcinomas and epithelial cancers. Lab invest 70(1):6-22.

2. Bonkhoff H, Remberger K. 1996. Differentiation pathways and histogenetic aspects of normal and abnormal prostatic growth: a stem cell model. Prostate 28(2):98-106.

3. Tsujimura A, Koikawa Y, Salm S, Takao T, Coetzee S, Moscatelli D, Shapiro E, Lepor H, Sun T-T, Wilson E.L. 2002. Proximal location of mouse prostate epithelial stem cells: a model of prostatic homeostasis. JCB 157(7):1257-1265.

4. Hayward SW, Haughney PC, Rosen MA, Greulich KM, Weier HU, Dahiya R, Cunha GR 1998. Interactions between adult human prostatic epithelium and rat urogenital sinus mesenchyme in a tissue recombination model. Differentiation 63(3):131-40.

5. Salm SN, Koikawa Y, Ogilvie V, Tsujimura A, Coetzee S, Moscatelli D, Moore E, Lepor H, Shapiro E, Sun TT, Wilson EL 2000. Generation of active TGF-beta by prostatic cell cocultures using novel basal and luminal prostatic epithelial cell lines. J Cell Physiol 184(1):70-9.

6. Salm SN, Koikawa Y, Ogilvie V, Tsujimura A, Coetzee S, Moscatelli D, Moore E, Lepor H, Shapiro E, Sun TT, Wilson EL. 2000. Transforming growth factor-beta is an autocrine mitogen for a novel androgen-responsive murine prostatic smooth muscle cell line, PSMC1. J Cell Physiol 185(3):416-24

7. Sugimura Y, Cunha GR, Donjacour AA 1986. Morphological and histological study of castration-induced degeneration and androgen-induced regeneration in the mouse prostate. Biol Reprod 34(5):973-83.

8. Lammie A, Drobnjak M, Gerald W, Saad A, Cote R, Cordon-Cardo C 1994. Expression of c-kit and kit ligand proteins in normal human tissues. J Histochem Cytochem 42(11):1417-25.

9. Crosby HA, Kelly DA, Strain AJ 2001. Human Hepatic Stem-like Cells Isolated Using c-kit or CD34 Can Differentiate Into Biliary Epithelium. 120(2):534-544.

10. Simak R, Capodieci P, Cohen DW, Fair WR, Scher H, Melamed J, Drobnjak M, Heston WD, Stix U, Steiner G, Cordon-Cardo C 2000. Expression of c-kit and kit-ligand in benign and malignant prostatic tissues. Histol Histopathol15(2):365-74.

The Location and Characterization of Murine Prostatic Stem Cells.

S. Salm [1], A. Tsujimura [1,2], Y. Koikawa [1,3], T. Takao [1,2], S. Coetzee[1], D. Moscatelli[1], T-T. Sun [1], E.L. Wilson [1]

1. New York University School of Medicine, New York, USA.
2. Osaka University Medical School, Osaka, Japan.
3. Fukuoka City Medical Center of Sick Children and Infectious Diseases, Fukuoka, Japan.

The characterization of prostatic stem cells is important for elucidating the mechanisms underlying the aberrant proliferation processes of benign prostatic hyperplasia and carcinoma. Since stem cells in other organs have been identified as slow cycling cells with a high proliferative potential, we used these features to locate the position of prostatic stem cells within murine prostatic ducts. We labeled the entire murine prostatic epithelial cell compartment with bromodeoxyuridine (BrdU). We then chased out the BrdU label by repeated cycles of prostatic involution and regeneration induced by the withdrawal and replacement of androgens. After each cycle, mice were sacrificed and their prostates examined immunohistochemically for BrdU.

We showed that after a 16-cycle chase period, 25% of the basal and luminal cells in the proximal region retained the BrdU label, whereas only 8% of the basal cells and 1% of the luminal cells in the distal region of the ducts were labeled. Thus the proximal region of the prostatic ducts is enriched with a subpopulation of basal and luminal epithelial cells that are label-retaining (thus slow cycling).

Culture of the proximal and distal cells revealed that the proximal cells have a high proliferative potential and are capable of reconstituting branched glandular ductal structures in collagen gels. Proximal cells gave rise to 10-fold more ducts in collagen than did the same number of distal cells. The ducts of proximal origin were 9-fold larger and had 8-fold more branches than those of distal origin. As proximal cells are slow cycling with high proliferative potential, we propose that the prostatic stem cell population is located in the this area.

Culture of the proximal cells in the presence and absence of various cytokines revealed that transforming growth factor-ß (TGF-ß) is inhibitory to their growth, reducing it by 20% at low concentrations (0.1 ng/ml). Addition of stem cell factor (SCF; 100 ng/ml) in conjunction with 0.5 ng/ml of TGF-ß "rescued" proximal cell growth by quenching the inhibitory effect of TGF-ß. Ablation of endogenous TGF-ß using a neutralizing antibody stimulated proximal cell growth by 28% over controls. Concurrent neutralization of TGF-ß and addition of SCF results in a 47% increase in growth. SCF alone does not result in a significant increase in growth.

We propose a model of prostatic homeostasis in which the prostate stem cells are located in the proximal region (adjacent to the urethra) of the prostatic ducts. These cells are maintained in a quiescent state by a balance between the inhibitory effects of TGF-ß and the stimulatory effects of cytokines such as SCF, in a manner similar to that described for hematopoiesis.

This work is supported by DoD grant DAMD17-02-1-0115 (Salm) and NIH grant DK52634 (Wilson).

The proximal region of murine prostatic ducts is enriched in prostate stem cells

Ken Goto, Sarah Salm, Sandy Coetzee, Herbert Lepor, Ellen Shapiro, David Moscatelli and E Lynette Wilson. Departments of Cell Biology and Urology, New York University School of Medicine, New York, USA.

The identification and characterization of prostatic stem cells is important as they may represent the target of carcinogenesis and the source of the abnormal proliferative events that accompany benign prostatic hyperplasia. Prostatic ducts consist of a proximal region attached to the urethra, an intermediate region and a distal tip. We have recently shown (J Cell Biol 157: 1257-1265, 2002) that the proximal region is enriched in a subpopulation of epithelial cells that have three important attributes of stem cells: they are slow cycling, possess a high in vitro proliferative potential and can reconstitute highly branched glandular ductal structures in collagen gels.

We therefore decided to investigate the growth properties of cells from the proximal and distal regions of murine ducts using an in vivo transplantation model. Cells were isolated by enzymatic digestion from the proximal and distal regions of ducts. They were combined with 18 day rat embryonic urogenital sinus mesenchyme (UGM) in collagen pellets (1×10^5 proximal or distal cells plus 2.5×10^5 UGM cells) and implanted under the renal capsule of adult male athymic mice. Tissue grafts were harvested after 8 weeks and weighed and processed for immunohistochemical examination.

There was a striking increase in the amount of tissue obtained after the implantation of proximal compared with distal cells. The mean wet weight of the tissue arising from the combination of proximal cells with UGM was 623 ± 183 mg compared with 45 ± 20 mg for the tissue obtained from the combination of distal cells with UGM ($p < 0.0001$). Minimal growth was noted with proximal, distal or UGM cells alone. Histological examination of the tissue showed the presence of abundant normal prostatic tissue comprised of ducts with a normal architecture consisting of basal and luminal cells. The lumens of many ducts contained prostatic secretory products indicating the presence of functionally normal prostatic tissue.

These experiments indicate that in an in vivo assay the proximal region of mouse prostatic ducts contains cells that have a greater proliferative potential than those obtained from the distal regions of ducts. This would be expected if this region contained large numbers of immature cells capable of regenerating functional prostatic tissue. These data provide additional compelling evidence that indicates that the proximal region of prostatic ducts is enriched in a stem cell population.

This work is supported by grants from the DOD (DAMD 17-02-1-0115) and the NIH (DK 52634).